



WATER RESOURCES RESEARCH GRANT PROPOSAL

Title: Microcystin-LR: a potential contaminant of concern for Iowa surface waters

Focus categories: TS, SW, NU

Keywords: microcystin, cyanobacteria, water quality, lakes

Duration: March 2000 – March 2001

Federal funds requested: \$16,246

Non-Federal funds pledged: \$33,000

Principal Investigator:

Maureen E. Clayton
Department of Biology
University of Northern Iowa
Cedar Falls, IA 50614-0421

Congressional District: 2

Statement of Critical Water Problems

Surface water quality is currently one of the most important environmental issues facing the state of Iowa, since the ecological, recreational and aesthetic values of these water bodies are threatened by non point source (NPS) pollution. The traditional threat to lake water quality from NPS contaminants is eutrophication resulting from nutrient loading. Preliminary evidence from a study of two Iowa lakes by a multidisciplinary team of researchers at the University of Northern Iowa, however, indicates that algal species composition may also be altered by nutrient loading. The dominance of cyanobacteria in eutrophic lakes suggests that water quality and human and wildlife health may be endangered by exposure to toxin producing strains of cyanobacteria. Several species of cyanobacteria, including species from the genera *Microcystis* and *Anabaena*, are known to produce biotoxins that are harmful to the health of humans and wildlife. The most toxic compound, microcystin-LR, is a potent inhibitor of protein phosphatases and is a tumor promotor. Both *Microcystis sp.* and *Anabaena sp.* have been seen as dominant components of Iowa lake phytoplankton. Therefore, we suggest that microcystin-LR may be a newly emerging contaminant of concern for surface water quality in Iowa.

Statement of Results or Benefits

The objective of the proposed research is to determine whether or not cyanobacterial biotoxins, in particular microcystin-LR, pose a significant threat to the quality of surface

waters in Iowa and to the health of humans and wildlife. We also propose to investigate the relationships among microcystin concentration, cyanobacterial populations, total phytoplankton populations, and nutrient concentrations in eutrophic Iowa lakes. The results of this study should indicate: (1) the degree of threat to ecosystem and human health, (2) correlations between toxin concentration and nutrient loading, and (3) an analysis of potential solutions to the problem, as well as directions for future research. We have developed a working relationship with the Iowa Department of Natural Resources, members of the Delaware and Blackhawk County Conservation Boards, and citizens concerned with the water quality of Lake Delhi and Silver Lake. We believe that the potential implications of this research project for ecosystem and human health risk will be of great benefit in discussions pertaining to the regulation of non point source pollution, not only in Iowa lakes but in all lakes potentially threatened by harmful algal blooms. The results of this study may also indicate the need to consider the need for and develop novel non-conventional mitigation actions for impacted lakes.

Nature, Scope and Objectives of the Research

Summer blooms of cyanobacterial populations are common in eutrophic lakes and ponds. These blooms represent very high concentrations of cyanobacteria in the water column, and may result in a discoloration of the water and in the formation of what is colloquially known as “pond scum”. Blooms are initiated and exacerbated by factors including excessive nitrogen (N) and phosphorus (P) loading, surface water temperatures $> 20^{\circ}\text{C}$, stratification of the water column, long water residence time, organic matter enrichment, low total and dissolved N:P ratios, and high irradiance and long daylength. These conditions are common in Iowa lakes and ponds, especially those which are impacted by nutrient loading from agricultural runoff.

High cyanobacterial concentrations may reduce recreational use of surface waters because of the aesthetic impact of the discolored water. In addition, some species of cyanobacteria produce biotoxins that can result in severe health impacts and death in humans and wildlife. Microcystin-LR is the most toxic of the cyanobacterial biotoxins; it is known to be a potent inhibitor of protein phosphatases 1 and 2A (Toivola et al 1994, Runnegar et al 1995, Sahin et al 1995, Craig et al 1996), as well as a tumor promotor (Lam et al 1995). Neurotoxic illness and liver failure have been observed in exposed humans (Jochimsen et al 1998, Pouria et al 1998). Microcystin-LR may also be transferred through the food chain (Kotak et al 1996, Prepas et al 1997) and can result in sublethal effects, including ionic imbalance, decreased grazing, and reduced growth, in fish and other aquatic organisms (Beveridge et al 1993, Keshavanath et al 1994, Bury et al 1995).

Species that are known to produce microcystin-LR (*Microcystis* sp. and *Anabaena* sp.) have been observed as dominant members of the phytoplankton assemblage in two Iowa lakes, Silver Lake and Casey Lake (Osterhaus 1999). Unfortunately, however, toxic and non-toxic strains are difficult or impossible to distinguish morphologically (Boney 1975). A direct competitive ELISA using antibodies against microcystin-LR (Lin and Chu 1994,

Liu et al 1996) is now available to quantitatively determine the microcystin concentration in surface waters.

We propose to use this ELISA to determine the degree of the potential threat posed by microcystin-LR to the health of the humans and wildlife (as well as the ecosystem) that utilize two Iowa lakes, Silver Lake (in the town of Delhi, Delaware County) and Casey Lake (in Hickory Hills Park, Tama County). These two lakes are under study in our laboratory because Silver Lake is heavily impacted by land use practices within the watershed (primarily agricultural) and does not support a recreational fishery, while Casey Lake has similar physical characteristics but different land uses (primarily parkland) and supports a thriving recreational fishery. The primary objective of the proposed research is to determine whether or not microcystin-LR should be considered as a contaminant of concern in surface waters.

Nutrient concentrations (N, P, K) will also be determined in the lakes. The nutrients N and P may stimulate cyanobacterial growth, while increased P is correlated with increased microcystin concentration within the cell (Rapala et al 1997); both pathways (increased cyanobacteria populations and increased toxin content per cell) will result in increased toxicity within the water column. Potassium (K) levels will be determined because potassium has been shown to suppress the growth of *Microcystis* (Parker et al 1997) and may therefore be a novel means of control of toxic cyanobacterial blooms.

Methods, Procedures, Facilities

Water samples will be collected weekly throughout the summer at stations within the body of Silver Lake and Casey Lake in conjunction with the UNI Lake Water Quality Study. Samples will also be collected from the permanent inflows to Casey Lake; Silver Lake has no permanent inflows and is fed by runoff. The samples will be transported to the laboratory on ice, stored at 4 °C, and processed as soon as possible.

Microcystin-LR concentrations will be determined by direct competitive ELISA (Lin and Chu 1994, Liu et al 1996). Toxin concentrations will be evaluated in relation to cyanobacterial species composition (determined by hemocytometer counting) and chlorophyll *a* levels (determined spectrophotometrically) as a measure of the total phytoplankton community. Concentrations of nitrogen, phosphorus, and potassium (K) will be measured by ion chromatography. Nitrogen and phosphorus will also be measured by colorimetric techniques to compare the accuracy of the two methods. With the exception of the microcystin ELISA, all of these techniques are routinely performed in our laboratory.

Related Research

Recent research on microcystin-LR has primarily focused on the following areas: (1) laboratory experiments to elucidate the mechanism of action (e.g. Toivola et al 1994, Lam et al 1995, Runnegar et al 1995, Sahin et al 1995, Craig et al 1996), (2) laboratory studies to examine the effects of nutrients on toxin production (e.g. Parker et al 1997,

Rapala et al 1997), (3) improved analytical methods for toxin detection (Lambert et al 1994, Lin and Chu 1994, Liu et al 1996, Ward et al 1997), and (4) mechanisms of toxin degradation (Tsuji et al 1994, Bourne et al 1996). This research suggests that cyanobacterial toxins may be environmentally relevant contaminants if toxin-producing strains are abundant, or if the strains are exceptionally toxic. The proposed research is relevant in that it will determine the potential threat from cyanobacterial toxins by determining the toxin content in surface waters and correlating these concentrations to cyanobacterial abundance and species composition, and to concentrations of nitrogen, phosphorus and potassium, physiologically important nutrients for cyanobacterial growth and toxin composition. The results of this research should provide information to assist resource managers in determining the extent of the problem and in suggesting the need for novel remediation strategies for eutrophic lakes.

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